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- L7 ANSWER 1 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
- TI Quaternary structure of rice nonsymbiotic hemoglobin.
- SO Journal of Biological Chemistry, (March 2, 2001) Vol. 276, No. 9, pp. 6834-6839. print.
 ISSN: 0021-9258.
- AU Goodman, Matthew D.; Hargrove, Mark S. (1)
- AB Plant nonsymbiotic hemoglobins are hexacoordinate heme proteins found in all plants. Although expression is linked with hypoxic environmental conditions (Taylor, E. R., Nie, X. Z., Alexander, W. M.,

and .

Hill, R. D. (1994) Plant Mol. Biol. 24, 853-862), no discrete physiological function has yet been attributed to this family of

proteins.

The crystal structure of a nonsymbiotic hemoglobin from rice has recently been determined. The crystalline protein is homodimeric and hexacoordinate with two histidine side chains coordinating the heme iron atom. Despite the fact that the amino acids responsible for the subunit interface are relatively conserved among the nonsymbiotic hemoglobins, previous work suggests that this group of proteins might display variability in quaternary structure (Duff, S. M. G., Wittenberg, J. B., and Hill, R. D. (1997) J. Biol. Chem. 272,

16746-16752;

Arredondo-Peter, R., Hargrove, M. S., Sarath, G., Moran, J. F., Lohrman, J., Olson, J. S., and Klucas, R. V. (1997) Plant Physiol. 115,

1259-1266).

Analytical ultracentrifugation and size exclusion high pressure liquid chromatography were used to investigate the quaternary structure of rice nonsymbiotic hemoglobin at various states of ligation and oxidation. Additionally, site-directed mutagenesis was used to test the role of several interface amino acids in dimer formation and ligand binding. Results were analyzed in light of possible physiological functions and indicate that the plant nonsymbiotic hemoglobins are not oxygen transport proteins but more closely resemble known oxygen sensors.

- L7 ANSWER 2 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 2
- TI A model for ligand binding to hexacoordinate hemoglobins.
- SO Biochemistry, (May 22, 2001) Vol. 40, No. 20, pp. 6155-6163. print. ISSN: 0006-2960.
- AU Trent, James T., III; Hvitved, Angela N.; Hargrove, Mark S. (1)
- AB Hexacoordinate hemoglobins are heme proteins capable of reversible intramolecular coordination of the ligand binding site by an amino acid side chain from within the heme pocket. Examples of these proteins are found in many living organisms ranging from prokaryotes to humans. The nonsymbiotic hemoglobins (nsHbs) are a class of hexacoordinate heme proteins present in all plants. The nsHb

from

rice (rHb1) has been used as a model system to develop methods for determining rate constants characterizing binding and dissociation of the His residue responsible for hexacoordination. Measurement of these reactions exploits laser flash photolysis to initiate the reaction from the unligated, pentacoordinate form of the heme protein. A model for ligand binding is presented that incorporates the reaction following

rapid

mixing with the reaction starting from the pentacoordinate hemoglobin (Hb). This model is based on results indicating that ligand binding to hexacoordinate Hbs is not a simple combination of competing first order (hexacoordination) and second order (exogenous ligand binding) reactions. Ligand binding following rapid mixing is a multiphasic reaction

displaying

time courses ranging from milliseconds to minutes. The new model incorporates a "closed", slow reacting form of the protein that is not at rapid equilibrium with the reactive conformation. It is also demonstrated that formation of the closed protein species is not dependent on hexacoordination.

- L7 ANSWER 3 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
- TI A model for ligand binding to hexacoordinate hemoglobins.
- SO Biophysical Journal, (January, 2001) Vol. 80, No. 1 Part 2, pp. 311a. print.

Meeting Info.: 45th Annual Meeting of the Biophysical Society Boston, Massachusetts, USA February 17-21, 2001 Biophysical Society . ISSN: 0006-3495.

AU Trent, James Thomas, III (1); Hvitved, Angela N. (1); Hargrove, Mark S. (1)

- L7 ANSWER 4 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC. DUPLICATE 3
- TI Nonsymbiotic hemoglobins in rice are synthesized during germination and in differentiating cell types.
- SO Protoplasma, (2001) Vol. 218, No. 3-4, pp. 125-133. print. ISSN: 0033-183X.
- AU Ross, E. J. H.; Shearman, L.; Mathiesen, M.; Zhou, Y. J.; Arredondo-Peter,

R.; Sarath, G. (1); Klucas, R. V.

AB Nonsymbiotic hemoglobins (ns-Hbs) previously have been found in monocots and dicots; however, very little is known about the tissue and cell type localization as well as the physiological function(s)

of these oxygen-binding proteins. We report the immunodetection and immunolocalization of ns-Hbs in rice (Oryza sativa L.) by Western

and in situ confocal laser scanning techniques. Ns-Hbs were detected in soluble extracts of different tissues from the developing rice seedling

immunoblotting. Levels of ns-Hbs increased in the germinating seed for the

first six days following inhibition and remained relatively constant thereafter. In contrast, ns-Hb levels decreased during leaf maturation. Roots and mesocotyls contained detectable, but low levels of ns-Hbs. Split-seed experiments revealed that ns-Hbs are synthesized de novo

seed germination and are expressed in the absence of any signal originating from the embryo. Immunolocalization of ns-Hbs by confocal microscopy indicated the presence of ns-Hbs primarily in differentiated and differentiating cell types of the developing seedling, such as the aleurone, scutellum, root cap cells, sclerenchyma, and tracheary

To our knowledge, this is the first report of the specific cellular localization of these proteins during seedling development.

- L7 ANSWER 5 OF 22 CAPLUS COPYRIGHT 2002 ACS
- TI Nonsymbiotic plant hemoglobins to maintain cell energy status in transgenic systems
- SO PCT Int. Appl., 44 pp. CODEN: PIXXD2
- IN Guy, Phillip; Duff, Stephen; Xianzhou, Nie; Hill, Robert; Durnin, Douglas;

Sowa, Aleksander

by

AB Nonsymbiotic Hbs are broadly present across evolution;
however, the function of these proteins is unknown. Cultured maize cells have been transformed to constitutively express a barley Hb gene in either

the sense (HB+) or antisense (HB-) orientation. Hb protein in the transformed cell lines was correspondingly higher or lower than in wild type cells under normal atm. conditions. Limiting oxygen availability,

placing the cells in a nitrogen atm. for 12 h, had little effect on the energy status of cells constitutively expressing Hb, but had a pronounced effect on both wild type and HB- cells, where ATP levels declined by 27% and 61%, resp. Energy charge was relatively unaffected by the treatment

in HB+ and wild type cells, but was reduced from $0.91\ \text{to}\ 0.73$ in HB-cells

suggesting that the latter were incapable of maintaining their energy status under the low oxygen regime. Similar results were obsd. with Pseudomonas aeruginosa cells transformed with an Hb expression vector.

is suggested that **nonsymbiotic Hbs** act to maintain the energy status of cells in low oxygen environments and that they accomplish

this effect by promoting glycolytic flux through NADH oxidn., resulting in

increased substrate level phosphorylation. Nonsymbiotic
Hbs are likely ancestors of an early form of Hb that sequestered
oxygen in low oxygen environments, providing a source of oxygen to
oxidize

NADH to provide ATP for cell growth and development. This in turn suggests that cells contg. increased levels of Hb protein will survive longer under low oxygen tension or high energy demand. Applications of transgenic Hb in improving agronomic properties of plants and in organ transplants are provided.

- L7 ANSWER 6 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
- TI A flash photolysis method to characterize hexacoordinate hemoglobin kinetics.
- SO Biophysical Journal, (November, 2000) Vol. 79, No. 5, pp. 2733-2738. print. ISSN: 0006-3495.
- AU Hargrove, Mark S. (1)
- AB A flash photolysis method is described for analyzing ligand binding to

new and growing group of hemoglobins which are hexacoordinate in the unligated, ferrous state. Simple analysis of a two exponential fit to time courses for CO rebinding at varying CO concentrations yields rate constants for formation and dissociation of the hexacoordinate complex, and the bimolecular rate constant for CO binding. This method was tested with a nonsymbiotic plant hemoglobin from rice for which these values had not previously been determined. For this protein, dissociation and rebinding of the hexaccordinating amino acid side chain, His73, is rapid and similar to the rate of CO binding at high CO concentrations. These results indicate that hexacoordination must be taken into account when evaluating the affinity of hexacoordinate hemoglobins for ligands.

- L7 ANSWER 7 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE 5
- TI Crystal structure of a nonsymbiotic plant hemoglobin.
- SO Structure (London), (15 September, 2000) Vol. 8, No. 9, pp. 1005-1014. print.
 ISSN: 0969-2126.
- AU Hargrove, Mark S. (1); Brucker, Eric Allen; Stec, Boguslaw; Sarath, Gautam; Arredondo-Peter, Raul; Klucas, Robert V.; Olson, John S.; Phillips, George N., Jr.
- AB Background: Nonsymbiotic hemoglobins (nsHbs) form a new class of plant proteins that is distinct genetically and structurally from leghemoglobins. They are found ubiquitously in plants and are expressed in low concentrations in a variety of tissues including roots and leaves. Their function involves a biochemical response to growth under

limited O2 conditions. Results: The first X-ray crystal structure of a member of this class of proteins, riceHbl, has been determined to 2.4 ANG

resolution using a combination of phasing techniques. The active site of ferric riceHbl differs significantly from those of traditional hemoglobins and myoglobins. The proximal and distal histidine sidechains coordinate directly to the heme iron, forming a hemichrome with

spectral properties similar to those of cytochrome b5. The crystal structure also shows that riceHb1 is a dimer with a novel interface formed

by close contacts between the G helix and the region between the B and C helices of the partner subunit. Conclusions: The bis-histidyl heme coordination found in riceHbl is unusual for a protein that binds O2 reversibly. However, the distal His73 is rapidly displaced by ferrous ligands, and the overall O2 affinity is ultra-high (KD apprxeq 1 nM). Our crystallographic model suggests that ligand binding occurs by an upward and outward movement of the E helix, concomitant dissociation of the distal histidine, possible repacking of the CD corner and folding of the

D helix. Although the functional relevance of quaternary structure in nsHbs is unclear, the role of two conserved residues in stabilizing the dimer interface has been identified.

- L7 ANSWER 8 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.
- TI Quaternary structure of nonsymbiotic plant hemoglobin.
- Biophysical Journal., (Jan., 2000) Vol. 78, No. 1 Part 2, pp. 166A. Meeting Info.: 44th Annual Meeting of the Biophysical Society. New Orleans, Louisiana, USA February 12-16, 2000 ISSN: 0006-3495.
- AU Goodman, Matthew D.; Fulton, D. B.; Hargrove, Mark S.
- L7 ANSWER 9 OF 22 CAPLUS COPYRIGHT 2002 ACS
- TI Nuclear localization of a hypoxia-inducible novel non-symbiotic hemoglobin

in cultured alfalfa cells

- SO FEBS Lett. (2000), 482(1,2), 125-130 CODEN: FEBLAL; ISSN: 0014-5793
- AU Seregelyes, C.; Mustardy, L.; Ayaydin, F.; Sass, L.; Kovacs, L.; Endre, G.; Lukacs, N.; Kovacs, I.; Vass, I.; Kiss, G. B.; Horvath, G. V.; Dudits,
- We have isolated a 483-bp-long full-length cDNA clone encoding a non-symbiotic Hb called Mhbl, the first one found in alfalfa. This non-symbiotic Hb is a single copy gene localized in linkage group 4 in diploid Medicago genome. The Mhbl mRNA was found only in the roots of alfalfa plants. The Mhbl gene was inducible by hypoxia and showed no induction by cold stress treatment. The Mhbl transcript level increased at the G2/M boundary in a synchronized alfalfa cell suspension culture. The majority of Mhbl protein was shown to be localized in the nucleus and smaller amts. were detected in the cytoplasm. A potential link to the nitric oxide signalling pathway is also discussed.
- L7 ANSWER 10 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
- TI Nonsymbiotic haemoglobins in plants.

6

- SO Acta Biochimica Polonica, (1999) Vol. 46, No. 2, pp. 431-445. ISSN: 0001-527X.
- AU Sowa, Aleksander W. (1); Guy, Phillip A.; Sowa, Slawomir; Hill, Robert D.
- AB General aspects regarding the presence of **nonsymbiotic haemoglobin** in plants are presented with the emphasis on those related to its function. As it becomes apparent that the **nonsymbiotic haemoglobins** are widespread across the

plant kingdom and that they represent a more primitive and evolutionary older form of the plant globin genes, the question of their function becomes more attractive. While the physiological functions of the symbiotic haemoglobins in plants are well understood, almost nothing is known about their nonsymbiotic predecessors.

Therefore, the known and hypothetical functions of haemoglobins in various systems are described along with information concerning properties and the regulation of expression of the nonsymbiotic haemoglobins. Interestingly, a number of nonsymbiotic haemoglobins have been shown to be hypoxia-inducible. The spatial and temporal pattern of this induction in barley may suggest that it is

an

in

integral part of the plants response to limiting oxygen stress.

- L7 ANSWER 11 OF 22 CAPLUS COPYRIGHT 2002 ACS
- TI Physiological studies on a **nonsymbiotic** plant hemoglobin in a transgenic maize cell system
- SO (1998) 223 pp. Avail.: UMI, Order No. DANQ32462 From: Diss. Abstr. Int., B 1999, 59(10), 5268
- AU Sowa, Aleksander Wladyslaw
- AB Unavailable
- L7 ANSWER 12 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
- TI Altering hemoglobin levels changes energy status in maize cells under hypoxia.
- SO Proceedings of the National Academy of Sciences of the United States of America, (Aug. 18, 1998) Vol. 95, No. 17, pp. 10317-10321. ISSN: 0027-8424.
- AU Sowa, Aleksander W.; Duff, Stephen M. G.; Guy, Phillip A.; Hill, Robert D.
 (1)
- AB Nonsymbiotic hemoglobins are broadly present across the plant kingdom; however, the function of these proteins is unknown. Cultured maize cells have been transformed to constitutively express a barley hemoglobin gene in either the sense (HB+) or antisense (HB-) orientation. Hemoglobin protein in the transformed cell lines correspondingly was higher or lower than in wild-type cells under normal atmospheric conditions. Limiting oxygen availability, by placing the cells
- in a nitrogen atmosphere for $12\ \mathrm{hr},\ \mathrm{had}\ \mathrm{little}\ \mathrm{effect}$ on the energy status
- of cells constitutively expressing hemoglobin, but had a pronounced effect
- on both wild-type and HB- cells, where ATP levels declined by 27% and 61%, $\,$
- respectively, Total adenylates in these cells were approximately 35% lower
 - than in HB+ cells. Energy charge was relatively unaffected by the treatment in HB+ and wild-type cells, but was reduced from 0.91 to 0.73
- HB- cells, suggesting that the latter were incapable of maintaining their energy status under the low oxygen regime. Treatment of the cells grown in
 - an air atmosphere with antimycin A gave essentially the same results. It is suggested that **nonsymbiotic hemoglobins** act in plants to maintain the energy status of cells in low oxygen environments and that they accomplish this effect by promoting glycolytic flux through NADH oxidation, resulting in increased substrate-level phosphorylation. Hypoxic acclimation of plants is an example of this effect in nature.

Nonsymbiotic hemoglobins are likely ancestors of an early form of hemoglobin that sequestered oxygen in low oxygen environments, providing a source of oxygen to oxidize NADH to provide ATP for cell growth and development.

L7 ANSWER 13 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE

Ω

TI Plant hemoglobins.

SO Plant Physiology (Rockville), (Dec., 1998) Vol. 118, No. 4, pp.

1121-1125.

ISSN: 0032-0889.

- AU Arredondo-Peter, Raul; Hargrove, Mark S.; Moran, Jose F.; Sarath, Gautam; Klucas, Robert V. (1)
- L7 ANSWER 14 OF 22 CAPLUS COPYRIGHT 2002 ACS
- TI Soybean (lbc3), Parasponia, and Trema hemoglobin gene promoters retain symbiotic and nonsymbiotic specificity in transgenic Casuarinaceae: implications for hemoglobin gene evolution and root nodule symbioses

SO Mol. Plant-Microbe Interact. (1998), 11(9), 887-894 CODEN: MPMIEL; ISSN: 0894-0282

AU Franche, Claudine; Diouf, Diaga; Laplaze, Laurent; Auguy, Florence; Frutz,

Thierry; Rio, Maryannick; Duhoux, Emile; Bogusz, Didier

AB Control of the expression of legume and nonlegume Hb genes was investigated. The Casuarina glauca and Allocasuarina verticillata transformation system was used to examine the properties of the soybean (1bc3), Parasponia andersonii, and Trema tomentosa Hb gene promoters in actinorhizal plants. Expression of the Hb promoters gus genes was examd. by fluorometric and histochem. assays. The fluorometric assays in

organs showed that the soybean and P. andersonii promoters were most active in nodules, whereas the T. tomentosa promoter gave a very high activity in roots. The histochem. study showed that GUS activity

by the soybean and the P. andersonii gus chimeric genes appeared mainly confined to the infected cells of the C. glauca and A. verticillata nodules. The T. tomentosa Hb promoter was primarily expressed in the root's cortex and vascular tissue. The results indicate that the soybean,

P. andersonii, and T. tomentosa Hb promoters retain their cell-specific expression in transgenic members of the Casuarinaceae, suggesting a close relationship between legume, Ulmaceae member, and actinorhizal Hb genes. The conservation of the mechanism for nodule-specific expression of soybean, P. andersonii, and C. glauca and A. verticillata Hb genes is discussed in view of recent mol. phylogenetic data that suggest a single origin for the predisposition to form root nodule symbioses.

- L7 ANSWER 15 OF 22 MEDLINE DUPLICATE 9
- TI A nonsymbiotic hemoglobin gene is expressed during somatic embryogenesis in Cichorium.
- SO BIOCHIMICA ET BIOPHYSICA ACTA, (1998 Nov 26) 1443 (1-2) 193-7. Journal code: AOW; 0217513. ISSN: 0006-3002.
- AU Hendriks T; Scheer I; Quillet M C; Randoux B; Delbreil B; Vasseur J; Hilbert J L
- AB After differential screening of a cDNA library corresponding to genes expressed during the early stages of somatic embryogenesis in leaf tissue from the Cichorium hybrid '474' (C. intybus L., var. sativumxC. endivia L., var. latifolia) a nonsymbiotic hemoglobin cDNA was obtained. Studies of the expression of the gene corresponding to this clone by

Northern blot analysis suggest that in Cichorium a nonsymbiotic hemoglobin gene is specifically expressed under somatic embryogenesis-inducing conditions, and that its expression is not related to stress caused by wounding or tissue culture conditions.

L7 ANSWER 16 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE

10

- TI Two hemoglobin genes in Arabidopsis thaliana: The evolutionary origins of leghemoglobins.
- SO Proceedings of the National Academy of Sciences of the United States of America, (Oct. 28, 1997) Vol. 94, No. 22, pp. 12230-12234.

 ISSN: 0027-8424.
- AU Trevaskis, Ben; Watts, Richard A.; Andersson, Carol R.; Llewellyn, Danny L.; Hargrove, Mark S.; Olson, John S.; Dennis, Elizabeth S. (1); Peacock, W. James
- AB We cloned two hemoglobin genes from Arabidopsis thaliana. One gene, AHB1, is related in sequence to the family of nonsymbiotic hemoglobin genes previously identified in a number of plant species (class 1). The second hemoglobin gene, AHB2, represents a class of nonsymbiotic hemoglobin (class 2) related in sequence to the symbiotic hemoglobin

of legumes and Casuarina. The properties of these two hemoglobins suggest that the two families of nonsymbiotic hemoglobins may differ in function from each other and from the symbiotic hemoglobins. AHB1 is induced, in both roots and rosette leaves, by low oxygen levels. Recombinant AHB1 has an oxygen affinity so high as to make it unlikely to function as an oxygen transporter. AHB2 is expressed at a low level in rosette leaves and is

low

temperature-inducible. AHB2 protein has a lower affinity for oxygen than AHB1 but is similar to AHB1 in having an unusually low, pH-sensitive oxygen off-rate.

L7 ANSWER 17 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE

11

- TI Rice **hemoglobins**. Gene cloning, analysis, and O2-binding kinetics of a recombinant protein synthesized in Escherichia coli.
- SO Plant Physiology (Rockville), (Nov., 1997) Vol. 115, No. 3, pp. 1259-1266.

ISSN: 0032-0889.

- AU Arredondo-Peter, Raul (1); Hargrove, Mark S.; Sarath, Gautam; Moran, Jose F.; Lohrman, Joseph; Olson, John S.; Klucas, Robert V.
- AB Although nonsymbiotic hemoglobins (Hbs) are found in different tissues of dicots and monocots, very-little is known about hb genes in monocots and the function of Hbs in nonsymbiotic tissues. We report the cloning and analysis of two rice (Oryza sativa L.) hb genes, hbl and hb2, that code for plant Hbs. Rice hbl and hb2 genes contain four exons and three introns, as with all of the known plant hb genes. At least three copies of the hb gene were detected in rice DNA, and

analysis of gene expression shows that hb1 and hb2 are expressed in leaves $% \left(1\right) =\left(1\right) +\left(1\right)$

but only hbl is expressed in roots. A cDNA for rice Hbl was expressed in Escherichia coli, and the recombinant Hb (rHbl) shows an unusually high affinity for O2 because of a very low dissociation constant. The absorbance spectra of the ferric and deoxyferrous rHbl indicate that, in contrast to symbiotic Hbs, a distal ligand is coordinated to the ligand-binding site. Mutation of the distal His demonstrates that this residue coordinates the heme Fe of ferric and deoxyferrous rHbl and

stabilizes O2 in oxy-rhbl. The biochemical properties of rice rHbl suggest $\,$

that this protein probably does not function to facilitate the diffusion of O2.

L7 - ANSWER 18 OF 22 MEDLINE

DUPLICATE 12

- TI Cell-specific expression of the promoters of two nonlegume hemoglobin genes in a transgenic legume, Lotus corniculatus.
- SO PLANT PHYSIOLOGY, (1997 Jan) 113 (1) 45-57. Journal code: P98; 0401224. ISSN: 0032-0889.
- AU Andersson C R; Llewellyn D J; Peacock W J; Dennis E S
- AB The promoters of the hemoglobin genes from the nitrogen-fixing tree Parasponia andersonii and the related nonnitrogen-fixing Trema tomentosa both confer beta-glucuronidase reporter gene expression to the central zone of the nodules of a transgenic legume, Lotus corniculatus. beta-Glucuronidase expression was high in the uninfected interstitial cells and parenchyma of the surrounding boundary layer and was low in the Rhizobium-infected cells. This contrasts with the expression of both the P. andersonii hemoglobin protein in P. andersonii nodules and the endogenous Lotus leghemoglobins that are expressed in the infected cells at very high levels. The expression pattern of the P. andersonii and T. tomentosa hemoglobin promoters in L. corniculatus resembles that of a nonsymbiotic hemoglobin gene from Casuarina glauca, which was introduced into this legume, and suggests that only the nonsymbiotic functions of the P. andersonii promoter are being recognized. Deletion of the distal segments of both the P. andersonii and T. tomentosa promoters identified regions important for the control of their tissue-specific and temporal activity in Lotus. Potential regulatory

elements, which enhance nodule expression and suppress nonnodule expression, were also identified and localized to a distal promoter segment. A proximal AAGAG motif is present in the P. andersonii, T. tomentosa, and nonsymbiotic Casuarina hemoglobin genes. Mutation of this motif in the P. andersonii promoter resulted in a significant reduction in both the nodule and root expression levels in L. corniculatus. Some of the regulatory motifs characterized are similar to, but different from, the nodulin motifs of the leghemoglobins.

- $\ensuremath{\mathsf{L7}}$ ANSWER 19 OF 22 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS INC.DUPLICATE
 - 13
- TI A new hemoglobin gene from soybean: A role for hemoglobin in all plants.
- SO Proceedings of the National Academy of Sciences of the United States of America, (1996) Vol. 93, No. 12, pp. 5682-5687.
 ISSN: 0027-8424.
- AU Andersson, Carol R.; Jensen, Erik Ostergaard; Llewellyn, Danny J.; Dennis,

Elizabeth S.; Peacock, W. James (1)

AB We have isolated a new hemoglobin gene from soybean. It is expressed in cotyledons, stems of seedlings, roots, young leaves, and in some cells in the nodules that are associated with the nitrogen-fixing Bradyrhizobium symbiont. This contrasts with the expression of the leghemoglobins, which are active only in the infected cells of the nodules. The deduced protein sequence of the new gene shows only 58% similarity to one of the soybean leghemoglobins, but 85-87% similarity to hemoglobins from the nonlegumes Parasponia, Casuarina, and barley. The pattern of expression and the gene sequence indicate that this new gene is a nonsymbiotic legume hemoglobin. The finding of this gene in legumes and similar genes in other species strengthens our previous suggestion that genomes of all plants contain hemoglobin genes. The specialized leghemoglobin gene family may have arisen from a preexisting

nonsymbiotic hemoglobin by gene duplication.

- L7 ANSWER 20 OF 22 MEDLINE DUPLICATE 14
- TI Symbiotic and nonsymbiotic hemoglobin genes of Casuarina glauca.
- SO PLANT CELL, (1995 Feb) 7 (2) 213-23.
 - Journal code: BJU; 9208688. ISSN: 1040-4651.
- AU Jacobsen-Lyon K; Jensen E O; Jorgensen J E; Marcker K A; Peacock W J; Dennis E S
- Casuarina glauca has a gene encoding hemoglobin (cashb-nonsym). This gene AB is expressed in a number of plant tissues. Casuarina also has a second family of hemoglobin genes (cashb-sym) expressed at a high level in the nodules that Casuarina forms in a nitrogen-fixing symbiosis with the actinomycete Frankia. Both the nonsymbiotic and symbiotic genes retained their specific patterns of expression when introduced into the legume Lotus corniculatus. We interpret this finding to mean that the controls of expression of the symbiotic gene in Casuarina must be similar to the controls of expression of the leghemoglobin genes that operate in nodules formed during the interaction between rhizobia and legumes. Deletion analyses of the promoters of the Casuarina symbiotic genes delineated a region that contains nodulin motifs identified in legumes; this region is critical for the controlled expression of the Casuarina gene. The finding that the nonsymbiotic Casuarina gene is also correctly expressed in L. corniculatus suggests to us that a comparable non-symbiotic hemoglobin gene will be found in legume species.
- L7 ANSWER 21 OF 22 CAPLUS COPYRIGHT 2002 ACS
- TI Organ regulated expression of the Parasponia andersonii hemoglobin gene

transgenic tobacco plants

SO MGG, Mol. Gen. Genet. (1988), 214(1), 68-73

CODEN: MGGEAE; ISSN: 0026-8925

- AU Landsmann, Jorg; Llewellyn, Danny; Dennis, Elizabeth S.; Peacock, W. James
- AB Plant Hb genes are known to occur in legume and non-legume families and in

both nodulating (e.g. P. andersonii) and non-nodulating species (e.g. Trema tomentosa). Their presence in non-nodulating plants raises the possibility that Hb might serve a function in non-symbiotic tissues distinct from their role in the nitrogen-fixing root nodules induced by micro-organisms. A P. andersonii Hb promoter was capable of expression

either the P. andersonii Hb gene, or a hybrid construct with the bacterial

chloramphenicol acetyltransferase gene (cat), in the nonsymbiotic plant, Nicotiana tabacum. Expression is predominantly in the roots, implying that Hb might have a function in roots of non-nodulated plants. A low level of Hb protein was also obsd. in non-nodulated P. andersonii roots, but not leaves, supporting this assertion. The expression in transgenic plants will allow further characterization of the promoter sequences essential for the organ-specific expression of Hbs in non-symbiotic tissues.

- L7 ANSWER 22 OF 22 CAPLUS COPYRIGHT 2002 ACS
- TI Common evolutionary origin of legume and non-legume plant hemoglobins
- SO Nature (London) (1986), 324(6093), 166-8 CODEN: NATUAS; ISSN: 0028-0836
- AU Landsmann, Jorg; Dennis, Elizabeth S.; Higgins, Thomas J. V.; Appleby, Cyril A.; Kortt, Alexander A.; Peacock, W. James
- AB The detection of **Hbs** in distantly related nonlegume plant families as well as in the legume families raises the question of whether

the origin of the plant Hbs was single or multiple. A Hb gene was isolated from Parasponia andersonii (Ulmaceae) which shows >50% nucleotide sequence homol. with the Hb genes of legume plants and has 3 introns at identical positions to the legHb introns. The Parasponia gene has homol. to Hb gene in another distantly related nodulating plant, Casuarina. It also hybridizes at high stringency to sequences in a related, but nonnodulating genus, Trema. Thus, the globin gene family

may

be widespread in modern plants; plant **Hbs** may have a cryptic function in **nonsymbiotic** tissue and plant **Hbs** have evolved by vertical descent, probably from an ancestor common to modern plants and animals.

=>

---Logging off of STN---

=>
Executing the logoff script...

=> LOG Y

COST IN U.S. DOLLARS	SINCE FILE	TOTAL
FULL ESTIMATED COST	ENTRY 45.06	SESSION 57.88
DISCOUNT AMOUNTS (FOR QUALIFYING ACCOUNTS)	SINCE FILE	TOTAL
CA SUBSCRIBER PRICE	ENTRY -3.10	SESSION -3.10